R126 33 D3 CMT

45. (New) The method of claim 29 wherein the ~~gram-negative bacterial~~ pathogen is of the genera ~~Haemophilus~~, Neisseria, Moraxella, Campylobacter, Shigella or Pseudomonas.

REMARKS

Applicant has carefully reviewed and considered the Office Action mailed on January 4, 2000, and the reference cited therewith.

Claims 22-23, and 29 are amended, claims 44 and 45 are newly added, and claims 30 and 31 are canceled; as a result, claims 22-26, 29, 44 and 45 are now pending in this application. No new subject matter has been added to the claims. The amendments to the claims are fully supported by the specification as originally filed. The amendments are made to clarify the claims, and are not intended to limit the scope of equivalents to which any claim element may be entitled.

A. Drawings

Formal drawings, satisfying the objections raised by the Reviewing Draftsperson, will be submitted to the Patent Office upon notification of allowance of the claims.

B. Non-Statutory Double Patenting Rejection

The Examiner rejected claims 22, 23, 25 and 29 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 19, 20 and 22 of U.S. Patent Application No. 08/565,943. Applicants will consider filing a terminal disclaimer upon notification of otherwise allowable subject matter. A terminal disclaimer may not be appropriate once the scope of allowable claims is determined in the present application.

C. 35 U.S.C. § 112, First Paragraph Rejection

The Examiner rejected claims 22-26 and 29 under 35 U.S.C. § 112, first paragraph, as failing to provide an enabling disclosure, because the specification does not provide evidence that the biological materials of the claimed invention are (1) known and readily available to the public; (2) reproducible from the written description, *e.g.*, sequenced; or (3) deposited. This rejection is respectfully traversed.

Upon receiving indication of allowable subject matter, Applicants will deposit plasmids pB28 and pB29 in compliance with the requirements set forth 37 C.F.R. 1.801-1.809.

The Examiner rejected claims 29 and 30 under 35 U.S.C. § 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventors, at the time the application was filed, had possession of the claimed invention. Claim 29 has been amended to indicate that the listed items are in the alternative; *i.e.*, the word "and" has been replace with the word "or." Claim 30 has been canceled.

D. 35 U.S.C. § 112, Second Paragraph Rejection

The Examiner rejected claims 22-26 and 29-31 under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter that Applicants regard as the invention.

The Examiner stated that claims 22 and 29 lack antecedence for the recitation "the" wild type gram-negative bacterial pathogen. Claims 22, 29 and 30 have been amended to recite "a" wild type gram-negative bacterial pathogen. Claim 30 has been canceled.

The Examiner stated that claim 23 lacks antecedence for "the *htrB* mutant". Claim 22 has been amended to provide proper antecedence for this term.

The Examiner stated that claims 29 and 30 are vague, confusing and/or incorrect in reciting three active ingredients in the vaccine formulation. Claim 29 has been amended to recite the active ingredients in the alternative. Claim 30 has been canceled.

The Examiner stated that claim 31 is unclear. Claim 31 has been canceled.

The Examiner stated that it is not clear what the differences are between claims 29 and 30. Claim 30 has been canceled, thereby rendering this rejection moot.

E. 35 U.S.C. § 102 Rejection

The Examiner rejected claim 22 under 35 U.S.C. § 102(b) as being anticipated by Karow et al. (J. Bacteriol. 174: 7407-7418, 1992). This rejection is respectfully traversed.

A proper rejection under §102(b) requires that the cited reference identically describe or disclose all of the elements of the claimed invention. The claims as amended recite a method of

making a mutant endotoxin lacking one or more secondary acyl chains of lipid A as compared to the wild type.

The present *htrb* mutant pathogen, such as in *H. influenzae*, makes a simple truncated penta- and tetraacylated lipid A, whose structure can be derived directly from the deletion of one or two O-linked myristoyl fatty acids (C_{14}) from the parental lipid A structure. § 132 Declaration of Drs. Gibson and Apicella, ¶ 4 (hereinafter "§ 132 Declaration", attached herewith). For example, in wild type strains, *Neisseria gonorrhoeae* lipid A is hexaacylated and contains two C-12 fatty acids (lauric acid), one on each of the two glucosamines. § 132 Declaration, ¶ 5. In contrast, the *htrB* mutation in *N. gonorrhoeae* strain 1291 results in the complete deletion of one of these two lauric acid moieties to form a pentaacyl lipid A structure. *Id.* No fully hexaacylated lipid A species is seen, nor higher mass structures or new fatty acids. *Id.* The outcome for *htrB* in *N. gonorrhoeae* is similar to the *htrB* knockout in *H. influenzae*, which produced a truncated pentaacyl and tetraacyl lipid A species. *Id.*

In addition, some changes in the phosphorylation pattern in the LOS and lipid A moiety are observed between wild type and *htrB*- mutant in *N. gonorrhoeae* strain 1291. § 132 Declaration, ¶ 6. These changes involve an increased level of phosphoethanolamine (PEA) in both the lipid A moiety as well as the oligosaccharide. *Id.*

The lipid A structures created by Karow *et al.* are quite different from those of the present invention. The present inventors obtained a culture of the *E. coli htrB* mutant (hereinafter "the Karow strain" or "the Karow mutant") from Costa Georgopoulos, one of the co-authors of the article Karow *et al*, J. Bacteriol. 174:7407-7418 (1992). §132 Declaration, ¶ 7. The present inventors then performed a mass spectrometric examination of the Karow strain. *Id.* The results of this examination clearly show that the Karow organism has a set of lipid A structures different in two very important ways from the *htrb* mutant pathogens of the present invention. *Id.*

First, the Karow mutant makes a fully hexaacylated lipid A structure that is distinct in mass from the lipid A made by the parental wild-type strain. § 132 Declaration, ¶ 8. Specifically, the Karow mutant appears to contain a mixture of new unsaturated fatty acids, most likely palmitoleic (C16:1) in place of the single lauric acid (C12:0) fatty acid. *Id.* This substitution causes a shift up in mass of 26 and 54 Da from the major wild type lipid A

(molecular weight = 1798), producing new hexaacylated lipid A molecules with molecular weights of 1824 (+26, or C_2H_2) and 1852 (+ 54, or C_4H_6). *Id.*

Second, even though pentaacyl and tetraacyl substituted lipid A species are also seen in Karow's *E. coli*, these structures, when present, are not simple deletions of one and two fatty acids from the wild type (as is the case for *H. influenzae htrB*), but rather contain at least one new fatty acid not present in the small amounts of corresponding pentaacyl lipid A (MW = 1588, wild type pentaacyl lipid A) seen in the wild type lipid A preparation. § 132 Declaration, ¶ 9. The molecular weights of these two lipid A molecules are 1616 and 1406, and are consistent with a loss of the palmitoleic group (-236 Da, MW 1852--> 1616, mutant pentaacyl lipid A) and then a myristic acid group (-210 Da, MW 1616--> 1406, mutant tetraacyl lipid A). *Id.*

Thus, significant differences exist in the lipid A structures in the *htrB* gene deletion mutants of the present invention as compared to Karow's strain. In particular, Karow's strain makes both a fully acylated lipid A as well as non-fully acylated lipid A, whereas the present invention contains only penta- and tetraacylated lipid A (*i.e.*, no fully acylated lipid A molecules). Therefore, the present invention is novel over Karow *et al.*

Not a limitation

F. 35 U.S.C. § 103 Rejection

The Examiner rejected claims 23-26 and 31 under 35 U.S.C. § 103(a) as being unpatentable over Karow et al. (J. Bacteriol. 174: 7407-7418, 1992) as applied to claim 22 above, and further in view of Gupta *et al.* (Infect. Immun. 60: 3201-3208, 1992).

Applicant respectfully submits that the Examiner has not established the *prima facie* obviousness of the present claims. To establish a *prima facie* case of obviousness, three basic criteria must be met. First, there must be some suggestion or motivation, either in the cited references themselves or in the knowledge generally available to an art worker, to modify the reference or to combine reference teachings so as to arrive at the claimed invention. *In re Fine*, 837 F.2d 1071, 1074 5 U.S.P.Q.2d 1596, 1598 (Fed. Cir. 1988). Second, the art must provide a reasonable expectation of success. *In re Vaeck*, 947 F.2d 488, 493, 20 U.S.P.Q.2d 14838, 1442 (Fed. Cir. 1991). Finally, the prior art reference must teach or suggest all of the claim limitations, and the teachings or suggestion, as well as the expectation of success, must come from the prior art, not applicant's disclosure. *Id.*

The motivation to modify the prior art must flow from some teaching in the art that suggests the desirability or incentive to make the modification needed to arrive at the claimed invention. *In re Laskowski*, 871 F.2d 115, 117, 10 U.S.P.Q.2d 1397, 1398 (Fed. Cir. 1989) ("[t]he mere fact that the prior art could be so modified would not have made the modification obvious unless the prior art suggested the desirability of the modification"). Karow *et al.* would not motivate the art worker to attempt to produce the claimed invention. Karow *et al.*, as discussed above, teaches the production of insertion mutations in the *E. coli htrB* gene that results in a strain that makes both a fully acylated lipid A (*i.e.*, hexaacylated lipid A) as well as non-fully acylated lipid A. In contrast, the present claims recite a pathogen that makes an endotoxin lacking one or more secondary acyl chains of lipid A (*i.e.*, only penta- and tetraacylated lipid A).

There is simply no teaching in Karow *et al.* to suggest to those skilled in the art to make a mutation that results in an endotoxin that has a decreased number of acyl chains. One certainly would not have had a reasonable expectation that such an endotoxin would have substantially reduced toxicity. Not only must one have a motivation to try to make the invention, there must also be a reasonable expectation of success. Further, both the suggestion and the reasonable expectation of success must be found in the prior art, not in the applicant's disclosure.

Gupta *et al.* does not remedy the deficiencies of Karow *et al.* Gupta *et al.* disclose the conjugation of chemically-modified LPS to cholera toxin and other proteins. They do not, however, teach or suggest a method of making an endotoxin that has a decreased number of acyl chains.

Therefore, the present invention is not obvious over Karow *et al.* in view of Gupta *et al.*

G. Objections

The Examiner objected to claim 30 as grammatically incorrect in the recitation "comprising an active ingredient an htrB mutant". Claim 30 has been canceled, thereby rendering this objection moot.

CONCLUSION

Applicant believes the claims are in condition for allowance and request reconsideration of the application and allowance of the claims. The Examiner is invited to telephone the below-signed attorney at (612) 373-6961 to discuss any questions which may remain with respect to the present application.

Respectfully submitted,

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By their Representatives,

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Date 30 June 2000 By _____

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I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to Box AF, Assistant Commissioner of Patents, Washington, D.C. 20231 on **June 30, 2000.**

Ann S. Viksnins _____

Name Signature